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INFECTIONS OF FETUSES AND FOALS

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BULLETIN NO. 333

(RESEARCH BULLETIN)

Infections of Fetuses and Foals

By W. W. DIMOCK and P. R. EDWARDS

From the data gathered on the causes of sterility in mares it has been determined that from 25 to 35 per cent of all non-producing mares, as they were found on the breeding farms in Kentucky, were barren because of pathological changes in the reproductive organs due to infection. The percentage of barren mares infected varied somewhat from year to year. In the beginning of the investigation only mares suspected of being infected were examined and the percentage found infected was high. Later, when it became the practice to examine all the barren mares on a farm, the percentage of mares examined and found infected naturally was reduced. Of the mares that showed infection of the genital tract in the form of cervicitis and metritis approximately two-thirds of the cases were due to streptococcic infection. It was found that a considerable number of the barren mares that were made available for examination and were under treatment had a history of abortion or that the last foal, altho carried full term, was either born dead or died early. Further, mares known to be suffering with inflammatory changes in the genital tract often were bred and of those that conceived a certain percentage either aborted or gave birth to diseased foals.

The objects of the investigation were: to determine the frequency of infection in aborted fetuses and foals; to obtain definite information on the particular types of microorganisms present and the relation, frequency, and importance of each; to ascertain the frequency of prenatal infection of foals and the

type of bacteria involved; to ascertain the relation of prenatal infection in foals to infections of the genital tract of mares and to breeding hygiene in the prevention of abortion and of diseases of the new born.

The problem of so-called contagious abortion in mares was not a part of this investigation. Abortion of mares due to *Salmonella abortivo-equinus* is recognized as a definite, independent entity. The microorganism is well established as a pathogen. Accurate methods of diagnosis are known and highly effective preventive measures have been worked out (1). Contagious abortion is to be considered in connection with abortion due to other causes only in the differential diagnosis and the care of the mare that has aborted. *S. abortivo-equinus* does not, as a rule, remain in the uterus of the mare and is only indirectly a factor in producing metritis following an abortion. Serious and chronic inflammatory changes that develop in the uterus of the mare following abortion due to *S. abortivo-equinus* are the result of other bacteria that may subsequently invade the uterus.

MATERIAL EXAMINED

It is extremely difficult, if not impossible, to draw a line between abortion and premature birth; therefore, it was necessary to set an arbitrary standard as to what specimens should be classified as fetuses and what should be called foals. We have designated as a fetus any specimen which was dead at delivery and in which parturition was two weeks or more premature. Any specimen which was living at the time of parturition, provided it was not more than thirty days premature, or was born dead within two weeks of the normal period of gestation, was classified as a foal.

The data on fetuses and foals were derived from the examination of 325 fetuses and 252 foals. The material was brought to the laboratory directly from the farms where the losses occurred and, with few exceptions, was received in condition satisfactory for examination. The placenta usually was available with the fetus altho often so badly torn and soiled as to render a

bacteriological examination of little value. Of the foals received, that were dead at birth, the placentas usually were available for study and often yielded valuable information. A number of placentas of mares that had normal gestation periods and delivered apparently healthy foals were studied. Information of value was obtained from examination of the genital tracts of mares following abortion, the birth of a diseased foal, or the birth of a normal foal. The method of examination of fetuses and foals was as follows:

At autopsy search was made for defects of development and visible tissue changes of a strictly pathological nature. Gross pathological changes are much more easily recognized and interpreted in foals than in fetuses. Special attention was given to gross lesions in foals of all ages. The location, extent and character of lesions observed were studied in order to learn whether it was possible or not to make from gross pathology alone a differential diagnosis that could be confirmed by bacteriological methods. It was found that only in the more typical and extreme cases and especially in the older foals was it possible to tell on gross examination just what type of infection was present. Altho it was not possible to tell what particular micro-organism was etiologically responsible for the lesions it was often possible to tell from gross examination which cases would prove to be positive or negative on bacteriological examination. It thus was determined that for an accurate diagnosis of disease of the new-born that terminates in death, bacteriological methods are required in the large majority of cases.

As a routine procedure in the examination of fetuses culture tubes were inoculated from the umbilical vessels, peritoneal cavity, pericardial sac, heart blood, liver, spleen, kidneys, lungs, and stomach contents. Only occasionally were culture tubes inoculated from other parts and structures. In the examination of the placenta culture tubes usually were inoculated from the umbilical cord, large blood vessels of both the amnion and chorion, and from the maternal surface of the chorion when contaminating material was not pronounced.

In the examination of foals culture tubes were inoculated

regularly from the peritoneal cavity, pericardial sac, heart blood, liver, lungs, spleen, kidneys, umbilical vein, and the joints. If the foal was dead at birth material was taken from the umbilical vessels and the stomach. In a few cases culture were made from the brain, cord, reproductive organs and other parts as conditions indicated. In a large percentage of foals examined the joint cavities proved the most satisfactory structure from which to obtain cultures. In a few cases *Shigella equirulis** was recovered in pure culture from the hip joint when cultures from all other joints were negative. In septicemic cases, including both streptococcus and *S. equirulis* infection, cultures from all parts usually were positive.

* Many names have been used for this species. The first was *Bacillus nephritidis equi*, by Meyer. The second was *Bacillus equirulis*, by van Straaten.

Magnusson called it *Bact. viscosum equi*. Bergey has recently classified the organism as *Shigella viscosa*. Since *Bacillus nephritidis equi* is a trinomial which in a strictly grammatical sense cannot be hyphenated, it seems that the valid name for this species is *Shigella equirulis* (van Straaten). The synonyms which have been encountered are as follows:

Bacillus nephritidis equi Meyer, Transvaal Dept. Agr., Rpt. of the Govt. Bacteriologist, 1908-9, 122-163.

Bacillus equirulis van Straaten, Verslag van de Werksaamheden der Rijksserumirichting, 1916-17.

Bact. viscosum equi Magnusson, Svensk. Veternärstidskr., 1917, 81.

Bacillus pyosepticus equi de Blieck u. van Heelsbergen, Tijdschr. v. Diergeneesk., 1919, 46, 492.

Bact. pyosepticum viscosum Miessner, Deut. tierärztl. Wchnschr., 1921, 29, 185.

Bact. pyosepticum viscosum equi Lütje, Deut. tierärztl. Wchnschr., 1921, 29, 185.

Bact. pyosepticum (viscosum) Miessner u. Berge, Deut. tierärztl. Wchnschr., 1922, 30, 473.

Bact. pyosepticum Miessner, Deut. tierärztl. Wchnschr., 1923, 31, 348.

Bact. pyosepticum equi Laudien, Inaug. Diss., Hannover, 1923.

B. pyosepticus Clarenberg, Ztschr. f. Infekkr. u. Hyg. d. Haust., 1924, 27, 193.

Bact. equi Weldin and Levine, Abstr. Bact., 1923, 7, 13.

Eberthella viscosa Snyder, Jour. Am. Vet. Med. Assoc., 1925, 66, 481.

Shigella equi Weldin, Iowa State Col. J. Sci., 1927, 1, 121.

Shigella viscosa (Magnusson) Snyder, Bergey, et al., Manual of Determinative Bacteriology, 3rd edition, 1930, 363.

During the ten-year period, 1921 to 1931, 325 aborted fetuses were brought to the laboratory for examination.

Following are the numbers and corresponding percentages of fetuses from which the various microorganisms were isolated.

Fetuses	Number	Percentage
Total	325	100
Negative	186	57.2
Negative (twins)	22	6.8
Streptococcus	86	26.5
<i>S. abortivo-equinus</i>	19	5.8
Staphylococci	6	1.8
<i>E. coli</i>	4	1.2
<i>S. equirulis</i>	2	.6

It will be noted that the percentage of *S. abortivo-equinus* infections was low (5.8%). This is partly because practically all the material was drawn from farms where vaccination against equine contagious abortion was practiced and partly because, where several outbreaks occurred, not all the aborted fetuses were presented for examination. After the diagnosis of contagious abortion had been made from an examination of some fetuses the later abortions in the same outbreak usually were confirmed as *Salmonella abortivo-equinus* infection by the agglutination test or by culturing the uterine excretion of the mares. In one outbreak of equine contagious abortion coming under our observation forty out of approximately 100 mares aborted but only six fetuses were examined. In two other outbreaks in small groups of mares not all the fetuses were presented for examination.

STREPTOCOCCIC INFECTION OF FETUSES.

Streptococci were isolated from 86, or 26.5 percent of the 325 fetuses examined. In many instances the organisms were isolated from all the internal organs of the fetuses. In others they were recovered from only one or two organs. In some

cases streptococci were present only on the cultures from the stomach fluid and lungs, indicating that they were present in the amniotic fluid. Often streptococci were present only on cultures from the umbilical cord and the blood vessels of the chorion. The maternal surface of the chorion frequently yielded streptococci when other parts of the membranes and the fetus were negative.

When streptococci are recovered from one or more organs of the fetus or from the umbilical vein and blood vessels of the chorion there can be no doubt that the organisms were present in the uterus of the mare. When streptococci are found only on the maternal surface of the chorion there is some question whether they may not be present from contamination of the membranes, since it is known that streptococci exist on the external genitals of the mare and the placenta may be soiled with feces and stable refuse. Experience has shown, however, that the soiling of the membranes does not result in the recovery of hemolytic streptococci when ante-partum infection is absent and that they are isolated only from placentas that exhibit gross evidence of pathological processes. Soiling of the membranes may mask the presence of streptococci thru gross contamination.

It is generally known that streptococci cause abortion thru their pathological action upon the chorion. Invasion of the fetus is a secondary phenomenon and may or may not occur. It is evident that in many cases of intra-uterine infection abortion occurs before the tissues of the fetus are invaded by streptococci. If the placentas were received with all the fetuses submitted and were in fit condition to permit a satisfactory bacteriological examination, streptococci probably would be found more frequently to be the etiologic agent in equine abortion.

Because of the frequency of cases in which the tissues of the fetus are bacteriologically negative, it becomes necessary to examine the placenta thoroly for evidence of disease and to take cultures from the placenta from every case when possible. It is also of great value in the differential diagnosis of abortion to know the condition of the genital tract of a mare that has aborted and her breeding record.

ABORTION DUE TO OTHER MICROORGANISMS.

Other bacteria occasionally encountered as infections in fetuses are *S. equirulis*, *Escherichia coli*, and staphylococci. *E. coli* and staphylococci often occur as contaminating organisms, but when they are recovered in pure culture from the organs of the fetus it is evident that they were present before abortion occurred. These microorganisms are only occasionally the cause of abortion. In previous work it has been demonstrated that bacilli of the colon group may become established in the genital tract of certain mares. When such mares conceive it is quite probable that colon bacilli remain active, produce gross pathological changes in the placenta, and cause the death and expulsion of the fetus. Our work also indicates that staphylococci and other semi-parasitic microorganisms, when present in the uterus, may produce changes that result in abortion.

S. equirulis has been found only rarely as a cause of abortion in mares, altho it is the microorganism most frequently found as a cause of infection in the newborn.

From our study of abortion in mares it is becoming more and more apparent that there are a number of cases in which the bacteria isolated, from the fetus, placenta, or genital tract of the mare that has aborted, have a direct etiological relation to the abortion. There are some cases of abortion that from the clinical history and pathological lesions appear to be bacterial in origin but from which no microorganisms are recovered. There are also many cases of abortion in mares that from every angle of approach are apparently not caused by the action of bacteria. While there are many individual cases of abortion in which no infection can be demonstrated, the more typical of the non-bacterial cases have been observed to occur among groups of mares on the same farm.

OUTBREAKS OF APPARENTLY NON-INFECTIOUS ABORTION.

Several well-defined outbreaks of abortion among mares, that were apparently not due to infection have been studied. A summarized description of three of the more typical will serve

to illustrate their general character. In the first, eleven out of twenty mares aborted. The mares were assembled in the early fall, housed in the same barn and pastured in the same field. They were well isolated from other horses, altho previous to the time of being brought together they had been in contact with other brood mares. Over one-half of the group were young mares in foal for the first time, the others were young but had previously produced foals. The feed for all was the same, consisting of hay and grain fed in the stable and corn stover fed in the field, on the ground. The mares had access to spring water in the field and city water in the stable. All were vaccinated against equine contagious abortion in October. Of the eleven mares that aborted two slipped on January 2, and one each on January 4, 5, 13, 15, 22, and 28, and February 10 and 20. On February 21st the ten which had not yet aborted were moved to another farm. The eleventh abortion occurred on February 23rd, the second day after the mares were moved. All the mares that aborted were well advanced in pregnancy, the majority being within three to six weeks of term. After the mares were moved feeding corn stover was discontinued and they no longer had access to spring water. The hay and grain fed, both before and after the mares were moved, was purchased on the open market.

Bacteriological examinations were made of all the fetuses. When routine cultures of the first three were negative, anærobic cultures and cultures on blood mediums were made of the remainder. These also were negative. Pregnant rabbits and guinea pigs were inoculated with suspensions of the grouped organs of several of the fetuses. The animals remained normal thruout a sixty-day period of observation. Every effort to demonstrate an infectious agent was negative. Feeding corn stover was the only variation from the usual method of feeding brood mares. At the time it was felt that possibly the corn stover was the medium from which some injurious agent was acquired. In subsequent outbreak of an apparently identical nature no corn stover was being fed. This tends to show that

the corn stover was not responsible or that other feeds may carry the same injurious factor.

The second clear-cut outbreak of apparently non-infectious abortion was among a group of thirteen mares. Nine of the thirteen aborted. These mares had been vaccinated previously against equine contagious abortion. The mares and fetuses were examined as in the previously described outbreak. In eight the results were negative. The ninth fetus was found to be infected with streptococci and the dam was found to be suffering with purulent streptococcic metritis. This mare may or may not have been affected as the other mares that aborted; however, it is probable that the streptococcic infection alone would have sooner or later produced abortion. The dams of the eight non-infected fetuses recovered promptly without treatment, were bred in the spring, conceived and gave birth to normal foals. The general health of the mares was not visibly affected before or after abortion.

These mares were stabled in the same barn and were being fed grain purchased on the local market and hay harvested on the farm. The hay was found upon examination to contain a considerable amount of pennyroyal (*Hedcoma pulegioides*). A second group of six pregnant mares on the same farm, but kept in a separate stable, were fed the same grain and water, cared for by the same attendants, pastured in the same field, but were fed shipped hay containing no pennyroyal. No abortions occurred in this group. Since the only difference in these two groups of mares was the hay which they were fed, the owner and the attending veterinarian were of the opinion that the hay was responsible for the abortion and that the pennyroyal was the direct cause of the trouble. Whether or not the pennyroyal was responsible for the abortions, no other cause was found. An ecbohic action has been attributed to certain volatile oils, among these oil of pennyroyal. The primary action of these oils is generally conceded to be that of a gastro-intestinal irritant, the ecbohic effect being secondary. None of the mares showed symptoms of gastro-intestinal disturbances.

The third outbreak of abortion in which no infection could be demonstrated occurred in a group of fourteen mares. Twelve of these were assembled in midsummer. They were housed in the same barn, received the same feed and water and were pastured in the same field. One of the twelve gave birth to a normal foal early in January. The first abortion occurred January 25th. Ten abortions then followed before another normal foal was born. The last abortion took place March 16. Following this three mares gave birth to normal foals. Two of the mares had been shipped to the farm from another state and aborted after they were on the farm 30 and 32 days, respectively. They were stabled with the other twelve mares for two days after their arrival at the farm but were never turned out with them. They were then moved to a distant barn where they later aborted. All the abortions occurred during the latter months of pregnancy, the fetuses being aborted from two to ten weeks before the mares were due to foal.

The mares and fetuses were examined as in the previous outbreaks. The results of the bacteriological examinations of both the mares and fetuses were uniformly negative. Pregnant rabbits and guinea pigs were inoculated with the blood and ground organs of several of the aborted fetuses. The animals remained normal and gave birth to normal young.

Three pregnant mares were used in transmission experiments. Two were fed 15cc each of mixed stomach fluid from two of the fetuses and injected intravenously with 5cc each of the mixed heart blood of the same fetuses. The third was injected subcutaneously with 10cc of the stomach fluid of a fetus and intravenously with 10cc of blood drawn from the juglar vein of the mare 24 hours following abortion. These mares were unaffected by the feeding and injections. Two of them gave birth to normal foals; the third, which had been purchased for the experiment, foaled 30 days prematurely, according to the breeding date given by the former owner. However, the foal was alive and altho small appeared normal and fully developed at birth. On the second day the foal showed symptoms of general weak-

ness and died within the next 48 hours. It was not delivered at the laboratory for examination until decomposition was well advanced. The lesions found were those of peritonitis and exudative pleurisy. *E. coli* was isolated from all internal organs and joint cavities.

None of the mares that aborted showed any clinical evidence of ill health before or following abortion. Two showed a purulent exudate following abortion but recovered promptly after treatment. The others made prompt recoveries without treatment. All were examined thirty days after the last abortion and cultures taken from the cervix were negative. All the mares were bred that year and 70 percent conceived.

In all these outbreaks the health of the mare was not visibly affected before or after abortion. Furthermore the get of two or more sires was represented in each of these outbreaks, thus eliminating the possibility that the trouble may have originated with the stallion.

RESULTS OF EXAMINATION OF FOALS.

During the ten-year period, 1921 to 1931, 254 foals were received for post-mortem examination. The foals varied in age from those dead at birth to 8 months of age. The majority were 24 hours to 1 week of age. Following are the numbers and percentages of the various types of infection encountered.

Total	Number	Percentage
Total	254	100
Streptococcus	65	25.6
<i>S. equirulis</i>	101	39.8
<i>E. coli</i>	11	4.3
<i>S. abortivo-equinus</i>	6	2.3
<i>Corynebact. equi</i>	5	2.0
Miscellaneous	11	4.3
Negative	45	17.7
Negative-Traumatic Injury..	5	2.0
Negative-Dystocia	5	2.0

If the age at death of the negative foals and those that died of infection due to streptococci and *S. equirulis* is considered some interesting facts are established. The number of foals of these three classes, that died at various ages, is given in table 1. The ages of two foals infected with streptococci, one infected with *S. equirulis* and one negative, are omitted because not known.

TABLE 1. Age of foals at death and kind of infection.

Age Days	Negative	Streptococcus infection	<i>S. equirulis</i> infection
1	38	14	36
2	1	4	11
3	2	2	8
4	1	3	6
5-7	1	6	6
8-14	4	9	2
15-30	3	9	4
over 30	4	16	27
Total	54	63	100

From table 1 it is apparent that most of the foals from which no microorganisms were isolated were dead at birth or died within the first day. The majority of this group were dead at birth. Among these are some known cases of dystocia. Probably many of these deaths were a result of difficult or delayed parturition, in which death is attributable to physiological causes. It is known that foals are more likely to die during difficult or delayed parturition than are the young of other species of domestic animals. Many foals in this group were found dead, no attendant being present at the time of birth. The majority of the foals in this group exhibited no pathological changes on post-mortem examination and it is the opinion of the writers that these deaths were not due to infection.

More than half the foals in which infection with *S. equirulis* was found, had died at the age of three days or less. Of foals

infected with streptococci not quite a third had died at that age. It is interesting that in the first week deaths due to *S. equirulis* greatly outnumbered deaths due to streptococci, whereas, during the second, third, and fourth weeks, deaths due to streptococci were three times as numerous as deaths due to *S. equirulis*.

In the group of foals that died at more than thirty days of age, the large majority were found to have a generalized infection. Microorganisms often become localized in the verminous aneurysms of the mesenteric arteries. Chronic infections in these lesions suddenly become active and terminate in septicemia.

STREPTOCOCCIC INFECTION OF FOALS.

Streptococci have long been known to be responsible for the death of foals. In the present work it was found that in 65 cases, or 25.6 percent, death of the foals was attributable to streptococcic infection. This infection is both prenatal and postnatal in origin. Cases of prenatal infection include foals dead or ill at birth and those that show symptoms of illness soon after birth. The latter usually died within 24 or 48 hours but occasionally lived for several days or weeks.

It will be noted from table 1 that 14, or 21.5 percent, of the foals infected with streptococci were dead at birth or died within 24 hours. These cases undoubtedly represent prenatal infection. From all the evidence obtainable it was apparent that the majority of streptococcic infections that culminated in death on the second and thru the 7th day, were also cases of intra-uterine infection. An acceptable history of all foals that showed streptococcic infection on examination was not often obtainable. In most instances when an accurate history could be obtained it was found that many of the foals that died as late as the 7th day had showed symptoms of illness at birth or shortly thereafter. Where fetal membranes from such cases were available examination revealed gross evidence of a diseased condition.

The symptoms and pathologic changes observed in foals that died from streptococcic infection varied with the age of the foal and the length of the illness. In foals born dead the changes

most often observed were petechial hemorrhages on the serous surfaces, particularly the pleura and epicardium. Peritonitis was present in some, and occasionally there were circumscribed areas of congestion and hemorrhage surrounding the umbilicus.

In the foals which were sick at birth and died within 24 to 48 hours the most prominent symptoms were extreme weakness and prostration. The most marked pathologic changes observed at post-mortem were hemorrhages of the serous surfaces, usually more numerous and more widely distributed than in the foals dead at birth. Inflammatory changes were evident in all the internal organs. Peritonitis was often present.

In foals which lived several days but which were not visibly ill when born the first symptom to become apparent was most often a disturbance of the digestive tract, diarrhea or constipation followed by diarrhea. The stump of the umbilical cord remained soft and oedematous. In some cases a purulent exudate escaped from the umbilicus. The temperature, respiration and pulse rate were increased and the appetite diminished. If the foal lived for several days after the appearance of symptoms, one or more joints of the legs usually became involved. Upon postmortem examination lesions of general septicemia were found in most cases. The umbilical vein was enlarged and in many cases filled with pus. The tissues immediately surrounding the umbilicus were inflamed and oedematous. In the majority of cases hemorrhages were general over the serous surfaces. Lesions in the lungs varied from congestion to inflammation and to well-developed pneumonic areas. Exudative pleurisy sometimes was pronounced. In some the liver was normal in appearance while in others it was enlarged, mottled, and showed changes varying from acute hepatitis to extreme inflammation and degeneration. The spleen usually was found to be somewhat enlarged and hemorrhagic. In the majority of cases the kidneys were found congested and sometimes definitely hemorrhagic. In cases of long standing there was usually a purulent arthritis. The arthritis involved the joint cavities and the surrounding tissues. Within the joint cavity the fluid was less viscous than normal, was

increased in amount, often blood-stained and in many instances purulent. It was not unusual to find that the joint cartilage was diseased and in many cases eroded.

In foals which did not show symptoms of illness before they were two to four weeks of age the disease either occurred as an extremely acute infection causing prostration and death within 24 hours or resulted in illness lasting several days to a week. In the first instance the only lesions apparent are those of general septicemia. In foals which were sick for some days before death gross pathologic changes of a more advanced type were evident in the internal organs and joint cavities.

CHARACTERS OF STREPTOCOCCI.

A detailed description of the streptococci concerned in genital diseases of the horse will not be given in this publication but a short characterization of this group is included in order that the reader may gain some idea of the etiological factor in the streptococcal infections which have been described. A detailed report of work with these organisms will be the subject of a future publication.

The streptococci found in infections of foals, as well as those recovered from aborted fetuses and from the genital tract of mares, correspond to the organism classified by Dimock and Snyder (2), Dimock and Caslick (3) and Dimock and Edwards (4) as *Str. genitalium* and to the organisms classified by Ogura (5) as *Str. pyogenes equi* A. They belong to the group of beta streptococci described by Brown (6). They cause a marked hemolysis both in blood agar plates and in liquid medium. In glucose broth they produce acidity rarely exceeding pH 4.9. Sodium hippurate is not hydrolyzed and methylene blue is not reduced. These streptococci ferment sucrose, lactose, and salicin and fail to attack mannite, inulin and rhamnose. They are highly virulent for laboratory animals.

From characters given above it is apparent that these organisms are members of the group classified by Brown, Frost, Shaw (7) as cultures of human origin. It was found possible,

however, to distinguish streptococci of human origin by their action on sorbitol and trehalose. The streptococci recovered from genital diseases in horses were found to produce acid from sorbitol but not from trehalose. The human strains, on the contrary, produce acid from trehalose but not from sorbitol. In a similar manner cultures from the genital tract can be distinguished from *Str. equi*; since Holth (8), Adersen (9), and Ogura (5) have demonstrated that *Str. equi* ferments neither lactose, sorbitol, nor trehalose.

S. EQUIRULIS INFECTION OF FOALS.

In any statement regarding the history, symptoms and pathology of infection due to *S. equirulis* the age of the infected individual must be considered. In a majority of foals that died of this infection death occurred on the first second or third day following birth. Practically all were visibly affected at birth. Many were so weak that they were unable to stand. Others were barely able to stand alone. Still others were able to stand and walk but were markedly sick, dull and listless. Some were not noticeably weak but showed symptoms of a nervous disorder, continually walking around the stall and refusing to nurse. In a few the first symptom was lameness. Others that appeared normal at birth were stricken suddenly and lived but a short time. The illness was characterized by sudden onset, extreme prostration, short duration after appearance of symptoms, and death. During the course of the disease the pulse, respiration and temperature were increased. Thirty-six percent of the foals which succumbed to the infection died within the first 24 hours. Such foals often exhibited no gross pathological changes. In some of them, however, an acute enteritis was apparent and occasionally peritonitis was observed. An acute nephritis was present and more rarely a purulent embolic nephritis involving the cortex of the kidney. The joints of foals that died during the first day were only very rarely visibly affected, altho in many instances the microörganism was present in the joint cavity.

In foals which died at two to three days of age the lesions

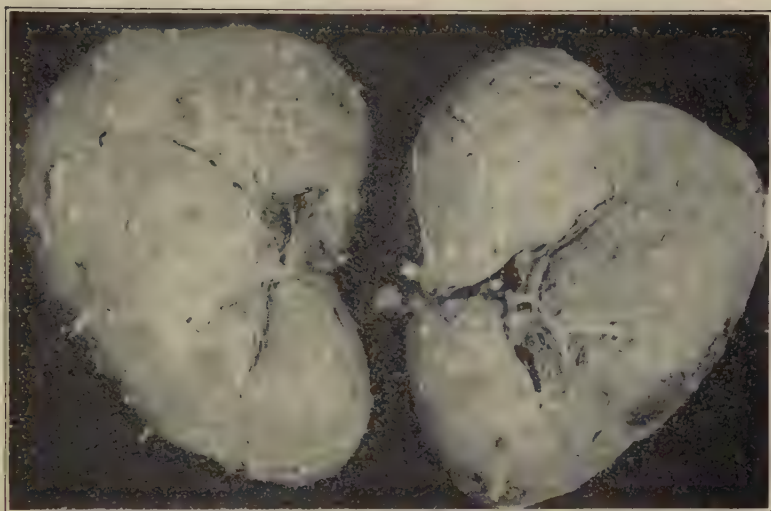


Fig. 1. Kidneys of four-day-old foal infected with *S. equirulis*, showing multiple abscesses.



Fig. 2. Section of kidney of four-day-old foal infected with *S. equirulis*, showing abscesses distributed thruout the cortex.

observed at post-mortem examination were more marked. These cases often exhibited a purulent nephritis, small abscesses being scattered thruout the cortex of the kidney. The size of these abscesses differed a great deal from one case to another, ranging from the point of visibility to 5 mm in diameter but in the same case they were remarkably uniform. There was a rather definite line of demarcation between the diseased area and the surrounding kidney tissue.

The distribution of the necrotic areas was consistently uniform with the glomerular structure of the kidney. This indicated that the condition was primarily a glomerulo-nephritis followed by suppuration and necrosis. Uniformity in size and distribution of the diseased areas indicated an independent origin and simultaneous distribution of the infection to each of the diseased foci in each individual kidney. In only one or two cases was a kidney found in which there was a noticeable difference in the size and distribution of the areas of necrosis, and in only two cases was a single, large, irregular, diseased area found. In one of the two latter cases (42-day-old foal) the diseased area was elliptical in outline and approximately 1 x 3 centimeters in size. It was at the margin of the renal hilus and extended from the surface thru the cortex well into the medulla. The diseased area was grayish white and had become well organized thru the ingrowth of connective tissue. In the other case (138-day-old foal) the lesion was in the anterior half of the right kidney, beginning at the rounded margin of the renal hilus and extending along the cortical portion to the renal sinus. It was in two rather distinct parts and at two points extended well down into the medullary portion. One part of the lesion was firm and fibrous; the other had the consistency of inspissated pus and showed a well-defined capsule. The lesion as a whole was very irregular in outline. It had average diameter of approximately four centimeters.

The joints of the legs were often affected. The joint lesions ranged from a slight increase in synovial fluid and a congestion of the joint capsule to a purulent arthritis involving the joint

cavity and tendon sheaths, with a great accumulation of fluid and extreme swelling. As a rule the more severe cases of arthritis were seen only in foals which were ill several days. The



Fig 3. Chronic lesion in kidney of 138-day-old foal infected with *S. equirulis*.

articular cartilages usually were normal. The joint capsule showed congestion even in the mildest cases. Further, the majority of the joints of all four legs showed evidence of disease to a mild or marked degree, the other extreme being that one joint showed purulent arthritis, while all the other joints of the legs appeared normal and were negative on culture. In mild cases the synovial fluid showed only a slight increase in viscosity and quantity and a somewhat darker color. In the more extreme cases a large amount of fluid was present. Upon opening the

joint cavity the fluid poured out with considerable force. The exudate varied in color from a light golden yellow to dark amber, and in those cases where the exudate was decidedly purulent in character it was a dirty brownish color. Air bubbles usually were present and gave the exudate a frothy appearance. It was always extremely viscid and tenacious and usually contained small flocculi; less often strings and tangled masses of coagulated mucus and fibrin.

Experience showed that the hock, hip and knee joints were more often involved than other joints of the legs. The tendon sheaths over the affected joint became greatly distended from the accumulation of a serous exudate. This was only slightly cloudy, containing a few pus cells. In a few cases this fluid was collected in the early stages of the disease, for bacteriological examination, but was negative on culture. Later, in the same foal, the exudate in the tendon sheaths and structures adjacent to the joint became purulent in character, of a very dark amber or brownish color, decidedly sticky and viscid, and proved positive on culture. It seems quite probable that these latter structures became involved thru direct invasion from the joint and not thru the general circulation.

A few foals showed an extreme purulent pleuro-pneumonia, the pleural cavity containing a large quantity of a purulent exudate. In cases that showed pneumonia, pleurisy or pericarditis, the organism was isolated from the structures of the pleural cavity in large numbers, but frequently was not recovered from any other part of the body. The symptoms and gross pathology observed in foals that died at ages of 4 to 7 days were much the same as in those that died in 2 to 3 days. The illness was usually of slightly longer duration and, for that reason, the pathological changes were more marked.

From Table 1 it will be seen that very few foals died of this infection between the ages of 7 days and one month, while quite a number died after reaching the age of 30 days. Most of these later cases were the result of infected verminous aneurysms of the mesenteric arteries. While a few of these older individuals

presented changes indicating infection at a very early age, such as chronic lesions of the kidneys, thick purulent exudate in the umbilical vein and inflammatory adhesions in the region of the umbilicus, in a greater number the predominating gross lesion was extensive purulent changes within verminous aneurysms in the mesenteric arteries.

When it is considered that practically every foal is affected with verminous aneurysms at an early age and that *S. equirulis* is frequently, if not constantly, present in the digestive tract of the horse, it is not surprising that such infections occur. *Strongylus* larvæ migrating from the intestine to the blood stream may serve as the agent for carrying many bacilli into the circulation of the parasitized individual. The larvæ migrate to the mesenteric blood vessels. The arteries, partially or totally occluded with fibrinous exudate and cell detritus, provide an area favorable for lodgment and development of microorganisms. Here a chronic infection develops which may suddenly become acute and result in septicemia and death.

These deductions are based not alone on pathological changes but, upon bacteriological examinations of infected aneurysms found in individuals that died of other causes (colitis, parasitism, ruptured intestines, streptococcic septicemia, etc.). *S. equirulis* has been found localized in these aneurysms when its presence could not be demonstrated in other parts of the body.

The disease in older foals was usually of a fulminating type, the hitherto apparently normal foal suddenly was prostrated and showed an extremely high temperature and greatly increased pulse rate. Death usually occurred in a few hours but in exceptional cases the animal lived for a week or more. The pathological changes encountered at post-mortem examination, as in the younger foals, depended to a great extent upon the length of the illness. Lesions of septicemia were generally present and in most cases there was a purulent nephritis with abscess formation. A purulent verminous aneurysm of the mesenteric artery was

almost invariably found. If the illness lasted for any length of time purulent arthritis usually developed.

HABITAT OF *S. EQUIRULIS*.

Laudien (10) demonstrated some years ago that *S. equirulis* was sometimes present in the intestinal tract of the horse. He succeeded in isolating the bacterium from intestinal contents in 13 instances. Jarmai (11) noted that septicemia due to *S. equirulis* in older foals and adult horses usually was preceded by a respiratory disturbance. From this he concluded that *S. equirulis* was a secondary invader which attacked the animals only when their resistance had been lowered by some other agent. He reasoned that *S. equirulis* probably was distributed very widely and was commonly present in the body of the normal horse. Working on this supposition he took cultures from the tonsillar area of 67 normal horses. In 49 of these cases he was able to isolate *S. equirulis*.

In the present work cultures were taken from the tonsillar region of 12 horses which were destroyed or which died from infection other than *S. equirulis*. In 10 of these cases *S. equirulis* was isolated and positively identified. The animals from which the organism was recovered varied in age from 17 days to 24 years. The 17-day-old foal died of streptococcic septicemia. Cultures from the tonsillar areas and from the taste buds yielded large numbers of *S. equirulis*. It is apparent that animals may become carriers of this microörganism at a very early age.

The bacterium has been found a number of times coexisting with streptococci in localized infections of horses, particularly in open abscesses and discharging sinues. A three day old foal was injected intratracheally with a culture of *Corynebact. equi*. On the ninth day following injection the animal developed pneumonia, became moribund, and was destroyed. *Corynebact. equi* was isolated from the lungs, the anterior mediostinal lymph gland, the liver and spleen. *S. equirulis* was present in the lungs but was recovered from no other organ. The foal had no

symptoms of *S. equirulis* infection and it is thought that the organism in this instance was a secondary invader.

Considering the almost constant presence of the bacterium in the mouth and pharynx of the horse, it is apparent that it would be present in the intestinal tract and feces. Hence any place where horses are kept would be contaminated with the organism. There is no definite knowledge of the viability of the bacterium in nature. It is possible that the organism is able to survive and multiply outside the animal body.

AGGLUTININS FOR *S. EQUIRULIS* IN BLOOD OF DAMS OF INFECTED FOALS.

It has been shown in an earlier publication (12) that different strains of *S. equirulis* differ markedly in their antigenic characters. It is obvious, therefore, that in any attempt to demonstrate specific agglutinins in the blood of mares that have given birth to infected foals, the homologous or infecting strain must be used as antigen. It must also be recognized that the serum of normal mares usually agglutinates *S. equirulis* in a dilution of 1 to 100, in many instances causes agglutination at 1 to 200, and rarely in higher dilutions.

The serums of 10 dams of infected foals were tested using the homologous organism as antigen. The mares were bled at periods varying from 2 to 10 days following foaling. In none of these tests was it possible to detect specific agglutinins for the infecting strain. In some instances marked agglutination was present in a dilution of 1 to 500, but when the same serum was titrated against heterologous strains known to be unrelated antigenically to the homologous strain, they were found to be agglutinated to the same degree.

RECOVERY OF *S. EQUIRULIS* FROM THE GENITAL TRACT OF DAMS OF INFECTED FOALS.

Since *S. equirulis* infection in foals was shown to be of pre-natal origin in some cases an attempt was made to recover the organism from the genital tract of mares whose foals died of

this infection. In all, 32 examinations were made on 24 mares. Only in two instances was *S. equirulis* isolated, once on the first day following foaling and once on the second day. These two cases furnish additional evidence that the disease was of prenatal origin. While it is believed that this infection often is contracted *in utero*, the writers are unable to explain the failure to recover the bacterium from the genital tract of the mare except in the two instances. Aside from the instances in which *S. equirulis* was isolated the results of the bacteriological examinations closely approximate those obtained in the examination of mares that had given birth to normal foals.

It is significant that mares rarely have two foals infected with *S. equirulis*. Only one mare of those under observation lost two foals from this infection. It is evident that the bacterium has no tendency to become established in the uterus. Also mares that have given birth to foals infected with this microorganism apparently have no greater tendency to develop metritis than those that have given birth to normal foals. Of the 24 mares reported upon here, 67 percent conceived and produced normal foals the following season.

TABLE 2. Examination of dams of foals infected with *S. Equirulis*.

Days after foaling	Negative	Streptococcus	<i>S. equirulis</i>	Coli	Total
1	2	2	1		5
2	1	2	1	1	5
3	1	2		1	4
4	2	2			4
5	1	1			2
6	1	1			2
7		1			1
8					
9		1			1
over 9	5	2		1	8
	—	—	—	—	—
	13	14	2	3	32

PATHOGENICITY OF *S. EQUIRULIS* AND ATTEMPTS TO REPRODUCE THE INFECTION IN HORSES.

S. equirulis is non-pathogenic for rabbits, guinea pigs and rats. Laboratory animals occasionally die after intraperitoneal or intravenous injection of large numbers of the bacterium but this action is apparently non-specific, since dead organisms are just as likely to produce death as living organisms. According to the reports of Lütje (13, 14) and Clarenburg (15) the organism is pathogenic for swine, both of these workers having observed cases of natural infection in pigs.

When *S. equirulis* is injected intravenously into foals 3 to 7 days of age it produces a rapidly fatal disease, the foal dying in two to four days with typical symptoms and lesions of the infection. As the age of the foal increases the resistance to artificial infection increases. In foals 6 to 10 months of age the bacterium rarely produces a fatal disease when large numbers of the organism are injected intravenously. Following the injection the foal is greatly depressed for several days. This depression is accompanied by increased temperature and pulse rate. Lameness develops and the joints of one or more legs are swollen. These joints often rupture and discharge a viscous, purulent exudate from which *S. equirulis* can be recovered in large numbers. These symptoms may persist for one to two weeks, after which the foal rapidly recovers. In some instances the swelling of the joints and lameness may recur periodically for 3 to 4 months and during these recurrences the bacterium can be recovered from the affected joints and from the subcutaneous tissue surrounding them.

Intravenous injection of the bacterium into older horses gives rise to symptoms closely resembling those produced in older foals but as a rule the changes are less severe and of shorter duration. Repeated intravenous injection of large numbers of the organism into adult horses may result in septicemia and death. This condition is exceptional, however; most adult horses gradually develop an increased tolerance to large doses administered intravenously. It should be stated that recently isolated

cultures of the bacterium are more virulent than old stock strains that have been maintained on laboratory mediums for long periods.

In attempts to produce *S. equirulis* infection as a prenatal disease in foals, pregnant mares were fed large numbers of recently isolated organisms from time to time during the period of gestation. The feeding was started early, as soon after breeding as pregnancy was assured, and continued periodically until the mare was within two weeks of foaling. These mares produced normal foals.

In two mares the organisms were introduced directly into the uterus two hours after service. The organisms were used in large numbers and were derived from recently isolated strains. These mares both conceived at the first service and produced normal foals. Examination of the placenta and the uterine exudate following parturition was negative. A third mare treated in the same way conceived at the first service but aborted from accidental infection with *S. abortivo-equinus*. As yet we have been unable to demonstrate the natural methods of infection in foals.

CHARACTERS OF SHIGELLA EQUIRULIS.

Since a detailed description of *S. equirulis* has been published (12), only a summary of the characters of this species will be given here. *S. equirulis* is a non-motile, non-sporing, Gram negative rod of variable morphology. Capsules have not been found. The bacillus produces ammonia in nitrate peptone broth and reduces nitrate to nitrite. It does not produce indol or hydrogen sulfide and fails to grow in uric acid or citrate mediums. The methyl-red and Voges-Proskauer tests are negative. Acid is produced in milk and some strains cause a coagulation of the casein. The fermentative properties vary slightly but as a rule dextrose, levulose, galactose, maltose, sucrose, lactose, xylose, raffinose, mannitol, glycerol, and dextrin are fermented with acid production, while arabinose, rhamnose, dulcitol, adonitol, salicin, inulin, and starch are not attacked. No gas production has been observed.

The organism, as it is usually isolated from infective material, produces rough, mucoid colonies. Rough and smooth, and mucoid and non-mucoid variation are extremely active in artificial cultures. In addition, dwarf-colony-forming strains sometimes are encountered. These dwarf colonies are small and transparent, and closely resemble colonies of streptococci. Since these different variants may be isolated directly from the tissues of infected foals, their recognition is of importance in the diagnosis of the disease.

In a study of a number of cultures of this species the antigenic characters of the organisms have been found to differ from strain to strain so that they form an extremely heterogeneous serologic group. Only a few instances of antigenic identity were established. For this reason it was impossible to develop serologic tests for the diagnosis of the infection or for the identification of the bacilli.

SOURCES OF INFECTION.

The origin of infection in the foals examined was not definitely determined for all cases. It is well established that this infection may be active in the pregnant uterus. Prenatal infection by *S. equirulis* was clearly demonstrated in cases of abortion and in foals dead at birth from which the organism was recovered. In foals that died within the first 24 hours the findings indicate prenatal infection. In those that died at 48 to 72 hours of age the evidence of prenatal infection cannot be taken as final; however, the foals were often weak at birth and showed other evidence indicating ante-partum infection. Of the foals that died at the age of 7 to 10 days some showed symptoms of illness at birth and were never normal.

It is the opinion of the writers that the majority of foals that died of *S. equirulis* infection within the first week were the victims of intra-uterine infection. In support of this view the following facts may be offered.

1. The infection was found in aborted fetuses and in foals dead at birth.

2. In a few instances the bacterium was demonstrated in the uterine exudate of the dam.

3. The presence of symptoms of infection at birth.

4. The extent, character and distribution of pathological changes.

5. Preventive measures at time of birth apparently are ineffective in the control of the disease.

In comparing the evidence of prenatal and postnatal infection it must be stated:

1. That the organism is commonly present in the digestive tract of the horse and therefore presumably present on objects with which the newborn foal comes in contact.

2. That in most instances the organism cannot be recovered from the uterine excretions of the dam.

3. Some foals 4 to 7 days of age, that yielded *S. equirulis* on post-mortem examination, are reported to have been normal at birth and showed no evidence of sickness until one to two days before death.

Adersen (quoted by Miessner and Wetzel (16)) has been widely quoted as having produced the disease by injection of cultures into the umbilical vein. This has been taken as evidence that the foal usually acquires the infection thru the umbilicus, after birth. When one considers that in this experiment 4cc of broth culture was injected directly into the umbilical vein it may be seen that the experiment amounted simply to intravenous injection. The conditions were in no way comparable to the exposure to which the foal is subjected in nature and the bearing of the experiment on the method of infection, is doubtful.

The statement that the infection attacks the foals of the same mare year after year is not, in our experience, applicable to this disease; it does apply to streptococcic infection. In the present work only one mare was known to have produced two foals infected with *S. equirulis*, and she produced a normal foal during the time that elapsed between the birth of the two diseased foals.

As has been previously stated, in foals six weeks of age or older, the organism is almost invariably recovered from verminous aneurysms of the mesenteric arteries; and in a few from this point only. It is quite probable that the larvæ of *Strongylus vulgaris* are the means by which the organism gains entrance. In two older foals lesions in the region of the umbilicus indicated that this was the portal of entrance and that infection had been present for some time. In two others a lesion in the kidney indicated infection of long standing.

The occurrence of *S. equirulis* infection in foals from two groups of mares is worthy of mention. As previously stated, 101 foals were found infected with *S. equirulis*. These were received from a large number of farms during a period of six years. With two exceptions, no more than two cases of this infection have been found on one farm the same year. The large majority of the infections occurred as individual cases. In one instance 4 cases occurred among a group of 12 mares, while in a second, 5 cases occurred in a group of 25 mares. In the group of twelve mares, two gave birth to dead foals which were 10 and 17 days premature, respectively, both bacteriologically negative. During the next fifteen days five mares foaled at term. One foal was dead at birth and was bacteriologically negative. The other 4 died at ages of 18 to 30 hours. All were infected with *S. equirulis*. The remaining 5 mares gave birth to normal foals. Other groups of pregnant mares on the same farm were unaffected.

In the group of 25 mares there were three abortions, the fetuses being from 30 to 80 days premature and bacteriologically negative. Three weeks following the third abortion a foal was born and died of *S. equirulis* infection at the age of 36 hours. Within a month four other foals died of this infection at ages varying from 14 hours to 7 days. Two mares then gave birth to dead foals which were negative on bacteriological examination. These were followed by a foal which died of streptococcic infection at the age of one month. The remaining 14 mares gave birth to normal foals.

Of the 9 cases of *S. equirulis* infection observed in these two

groups, 8 died at ages ranging from 14 to 48 hours. The ninth foal was 7 days of age. The early age at which the foals died indicates that these were cases of prenatal infection.

It is of interest that in these two groups the history is quite similar. The mares of both were affected with a rather mild febrile disease six weeks to two months before foaling. The symptoms were congested and icteric membranes, increased temperature, general depression and loss of appetite. In both groups, several abortions occurred before the diseased foals were born. No microorganisms were isolated from the fetuses. The mares that gave birth to diseased foals appeared extremely nervous and weak following parturition. In both groups the history of the mares, the occurrence of abortion and the loss of foals from *S. equirulis* infection were very similar. With the exception of the occurrence of the disease in these two groups of mares, all of the observed cases of the infection were individual and isolated.

INFECTION OF FOALS WITH *CORYNEBACTERIUM EQUI*.

Corynebacterium equi was first isolated and described by Magnusson (17) who found the organism in twelve cases of purulent pneumonia of foals. These cases were confined to one breeding establishment in southern Sweden. It was later found by Miessner and Wetzel (18), Lütje (19) and Lund (20) in pneumonias of foals in Germany. The first report of the occurrence of this infection outside of continental Europe was made by the writers (21) in 1930. The first case of this disease encountered was in a 42-day old foal brought to the laboratory for post-mortem examination on May 2, 1927. Since that time four other foals have been found to be suffering with this infection.

Symptoms. The disease usually occurs in foals 2 to 4 months of age. There is no apparent connection between this disease and the joint ills and navel ills of very young foals. In the majority of the cases reported the affected individuals had suffered no previous illness. The onset of the disease was

characterized by dullness, increased temperature, rapid pulse and respiration. The temperatures remained above normal during the course of the disease, sometimes exceeding 106°. A cough developed and the eyes became watery. There was a purulent discharge from the nasal passages. Moist rales developed in the lungs. The foal became weak and emaciated and death followed. The course of the disease was found to vary from 6 to 14 days. Our experience with the infection is too limited to make any estimate of the incidence or the mortality. Magnusson (17) reports a fatality of 80 percent.

Gross Pathology. In the majority of cases the disease apparently originated in the lungs. The most constant lesion was a broncho-pneumonia with multiple abscess formation. The abscesses were circumscribed and varied from 10 mm. to 100 mm. in diameter. They were scattered thruout the entire lung but were most numerous in the ventral portion. The abscesses were filled with a purulent, yellowish gray exudate. While the content of the smaller abscesses was usually fluid, in the larger ones it was more often inspissated. The lymph glands of the pulmonary region almost always were involved. The mediastinal glands were greatly enlarged and abscessed. In many cases the mesenteric, colic and hepatic lymph glands were greatly enlarged and contained abscesses. In some, multiple abscesses of the liver were observed. The abscesses in the lungs appeared to be the oldest lesions. The infection apparently became established in the lung tissue and spread thruout the body thru the lymphatics, and a septicemia eventually developed.

Morphological and Cultural Characters. In exudate from the abscesses and smears from agar slant cultures *Corynebact. equi* appeared as a short rod with rounded ends. It measured 1.2 microns x 1.5 microns. Many coccoid forms were seen. In broth cultures the morphology was quite different, the culture being made up of long rods and filaments. Numerous instances of branching were observed in broth cultures but not in smears from agar slants. The organism was non-motile and did not produce spores. Capsules were observed in moist India ink preparations. It was strongly gram positive.

The organism grew well on ordinary nutrient agar. Agar colonies attained a size of 1 to 3 mm. in 24 hours and after 48 hours incubation attained a diameter of 5 to 7 mm. The colonies were moist, slightly raised, round and entire. While the growth was not mucoid it was so moist that it streaked down an inclined surface. Thus a colony developing at the top of an agar slant runs down the slant during incubation causing a streak of growth. When grown in broth the organism produced an even clouding of the medium with little sediment and no pellicle formation.

The organism produced a yellowish-tan to salmon-pink pigment. The production of pigment was more pronounced on some media than on others. Indol was not formed, H_2S was not produced, and gelatin was not liquefied. Litmus milk remained unchanged. Nitrate was vigorously reduced to nitrite. Ammonia was not produced in nitrate peptone broth. On Loeffler's serum there was a light to moderate growth, less abundant than on agar. There was little pigment production on this medium. On potato a light growth occurred and a moderate amount of tan pigment was produced. If incubation was continued for 10 days secondary colonies appeared on the primary growth. These colonies were .5 to 1 mm. in diameter and produced a bright orange pigment, much deeper in color than that of the original culture. On coagulated egg yolk a vigorous growth took place. The growth was dryer than that on agar or Loeffler's serum. On this medium the pigment production was most pronounced, the growth being salmon pink. After two weeks incubation, the growth on egg yolk appeared dry, raised, and greatly wrinkled, resembling in many respects the growth of tubercle bacilli.

The organism was tested for its ability to produce acid and gas from glucose, levulose, galactose, lactose, sucrose, glycerol, mannitol, sorbitol, arabinose, xylose, raffinose, maltose, adonitol, salicin, inulin, dextrin, and starch. No acid nor gas production was noted in any of the tests.

Pathogenicity. *Corynebact. equi* apparently was non-pathogenic for the laboratory animals. Rats and guinea pigs were not affected by large doses administered intraperitoneally. Rab-

bits were injected intravenously with 1cc of broth culture. The animals remained normal and continued to gain weight during a 60-day period of observation.

Old horses were injected intravenously with 1 agar slant culture of the organism. The injection was followed by an immediate rise in temperature, increased rate of pulse and respiration, dullness and loss of appetite. The temperature reached 106°F. After 48 hours the temperature began to decline and the horse returned to normal. Subcutaneous injection of the bacterium in horses gave rise to local swellings that were hot and painful, followed by abscess formation. The abscesses ruptured and discharge a thick, yellowish pus.

Magnusson (17) reported the reproduction of the disease in a 3-months-old foal by intratracheal injection of the organism. The writers were unable to reproduce the condition by injection of cultures into the mouth and nasal passages. Two foals aged 2 months and 3 months were used. Large amounts of culture were injected into the upper nasal passages with a long-nozzle syringe. At the same time suspensions of the organism were injected into the throat. The foals remained healthy and there were no apparent ill effects from the exposure.

A three-day-old foal was injected intratracheally with one agar slant culture of the bacterium suspended in 2cc of saline. For four days following the injection the foal remained normal. On the fifth day it seemed weak and depressed. This depression was accompanied by intestinal disturbances which lasted until the animal died. On the fifth day the animal became weaker and moist rales developed in the lungs. Weakness increased and the foal became emaciated until on the ninth day the animal could neither stand or nurse. At this time the foal was destroyed. The temperature had remained normal during the course of the illness.

On post-mortem examination the lungs were found congested and pneumonic. No other lesions were observed. *Corynebact. equi* was recovered from the lungs, anterior mediastinal lymph gland, liver and spleen. Other cultures were negative. *S. equirulis* and streptococci were present in cultures from the

lungs. No exotoxin was demonstrated in filtrates of broth cultures of the bacilli.

Serological Reactions. Magnusson (17) reported that he was unable to produce agglutinins in rabbits by repeated intravenous injections. The writers produced agglutinating sera by the intravenous injection of horses. The animals were given 4 injections at intervals of 7 days. Each of the first two injections consisted of the growth from 1 agar slant culture. The last two were the growth from two agar slant cultures, each seven days after the last injection the animals were bled. At this time the serum had a titre of 1:100. Normal horse serum fails to agglutinate the organism. Insufficient work has been done to justify any conclusions as to the serological characters of the bacterium.

INFECTION OF FOALS BY MEMBERS OF THE COLON-AEROGENES GROUP.

In 11, or 4.3 percent, of the foals examined the only bacteria isolated were members of the colon-aerogenes group. In three cases the organisms in question were isolated from all the internal organs and in many cases from the joints. While there was always the possibility that bacilli of this type were present as a result of post-mortem invasion, in most of the instances recorded here they were the etiological factor of the disease. This view was supported by the fact that the animals were examined as soon after death as possible and in these cases there was no evidence of post-mortem decomposition.

These infections usually occurred in very young foals, the animals rarely being more than 48 to 72 hours old at death. Some of the foals were visibly ill at birth and died within a few hours. Others showed no symptoms of illness before they were 48 hours old. The animals had an increased temperature and pulse rate, were dull and weak and refused to nurse. Death usually occurred within 24 hours after the appearance of symptoms.

The changes observed at post-mortem examination were those of a general septicemia. Peritonitis was present. There

was often a severe enteritis. In many instances the joints were involved and the synovial fluid contained purulent flocculi.

It is believed that these infections are usually of uterine origin. In support of this view it may be said that these foals were often visibly ill at birth or shortly thereafter. In addition it is known that many mares are affected with metritis due to bacilli of this group. Some of these infections in foals undoubtedly originated from such mares.

FOALS BACTERIOLOGICALLY NEGATIVE.

No bacteria were isolated from 55, or 21.6 percent, of the foals examined. From table 1 it may be seen that 38 or 70% of the negative foals were born dead or died during the first day. The very large majority of these were dead at birth. Some of these deaths were known to be due to dystokia. In other cases there was a history of difficult or delayed parturition. It is probable that the death of most of these foals was due to physical causes. It is possible that a small number were the result of streptococcic infection but from which the microorganisms were not isolated.

The negative foals that died at ages of two or more days were sometimes found to have abnormalities of the internal organs, particularly the persistence of the ductus arteriosus and the foramen ovale and anomalous imperforations of the large and small intestines. Intussusceptions were also occasionally encountered. A few cases of rupture of the intestines were found. These usually occurred in heavily parasitized foals. Several older foals have been examined in which death was apparently due to colitis which was parasitic in origin. These cases, as a rule, were bacteriologically negative.

When these facts are taken into consideration only a small number of deaths remain unexplained. There were, however, a number of cases, largely among the foals dead at birth, in which no adequate explanation of the cause of death can be offered.

**EXAMINATION OF THE GENITAL TRACT OF MARES FOLLOWING
NORMAL PARTURITION.**

In the course of this investigation clinical and bacteriological examinations were made of the genital tract of 128 mares, following the birth of normal foals. This work was done in order to observe the clinical changes that normally take place following parturition, and to gain some idea of the uterine flora during the first nine days after foaling. Some of the mares were examined only once, others were examined repeatedly. The total number of bacteriological examinations was 220. The first examination of each mare was made between the date of parturition and the ninth day following; that is, between foaling and the first heat period. Several mares which were not bred on the ninth day, or which failed to conceive after having been bred on this heat period, were examined later.

In table 3 the bacteriological results are summarized. The findings on each day up to the ninth are given separately, while those made after the ninth day are grouped together. It is seen that the only organisms isolated with any degree of regularity were streptococci. These were present from the day following foaling up to the ninth day. It is seen that the percentage of negative results increased during the latter part of this period, probably due to the natural bactericidal power of the uterus and the outflow of the uterine secretions as œstrum approaches. In the examinations made after the ninth day the percentage of cultures showing streptococci was high. This is accounted for by the fact that these were mares which did not make normal recoveries after foaling, and many of them showed clinical evidence of metritis.

Streptococci were found in 72 percent of the fourteen mares examined on the first day following foaling. These findings demonstrate how early streptococci appear in the uterine secretions and coincide with the clinical history of the majority of cases of genital infection; that is, that streptococcic metritis usually originates soon after the mare has foaled. These results further show that uncertainty of efforts to demonstrate the etiology of stillbirths and abortions by bacteriological examina-

tion of the genital tract. Unless a specific microorganism, such as *S. abortivo-equinus*, is found, the results are not diagnostic. It should be stated in this connection that the streptococci isolated from mares after normal parturition were identical with streptococci found in metritis, abortion, and septicemia of foals.

Colon bacilli were occasionally isolated from mares after foaling. These were usually present during the early part of the period following foaling and tended to disappear before the ninth day. They were usually present with streptococci, rarely being found alone. In table 3 they are recorded only in those instances in which they were found alone. Other organisms such as micrococci, chromogenic rods, and actinomyces were rarely encountered.

Of the 128 mares examined, 85 showed streptococci on culture at some time following foaling; thirty-nine gave only negative cultures, and 4 gave only miscellaneous microorganisms (micrococci, chromogenic rods, etc.). While it was not possible to obtain the subsequent breeding records of all the mares, those which are known were as follows: Of the mares that showed

TABLE 3. Bacteriological examination of mares after foaling.*

Days	Negative	Streptococcus	E. coli	Miscellaneous	Total
1	3	10		1	14
2	6	8	3		17
3	6	20	1	1	28
4	13	18	2		33
5	9	8	1		18
6	4	7			11
7	11	9		1	21
8	7	7			14
9	4	2		1	7
over 9	21	35		1	57
	—	—	—	—	—
Total	84	124	7	5	220

* The mares represented in this table foaled normally at term and the foals were normal, healthy individuals.

streptococci at some time following foaling, 55% conceived that year. Two of these pregnancies were known to have terminated in abortion. Of the mares which did not yield streptococci on culture, 70% conceived that year and so far as known all these pregnancies resulted in normal births and healthy foals.

EXAMINATION OF THE GENITAL TRACT OF MARES FOLLOWING ABORTION.

Clinical and bacteriological examinations were made of the genital tract of 89 mares after abortion. Some of these were examined only once, others were examined repeatedly. The results of the bacteriological examinations are given in table 4.

It will be noted that *S. abortivo-equinus* was recovered only ten times. As stated earlier, this is accounted for by the fact that most of the material used in this study was drawn from farms practicing vaccination against equine contagious abortion. With the exception of the isolation of *S. abortivo-equinus* from

TABLE 4. Bacteriological examination of mares after abortion.

Days	Negative	Streptococcus	<i>S. abortivo-equinus</i>	<i>E. coli</i>	Miscellaneous	Total
1	8	9	1	1	1	20
2	4	10	2			16
3	2	5	1			8
4	1	3			1	5
5	1	4	2			7
6	2	3	1			6
7	1	1				2
8	1	4				5
9	3	2				5
over 9	24	16	3		1	44
Total	47	57	10	1	3	118

these 10 mares there was very little difference in the uterine flora of mares that had aborted and of those that had given birth to normal foals. Certain mares were negative on bacteriological examination, but the majority yielded streptococci at some period following abortion. Unless a specific microörganism such as *S. abortivo-equinus* is isolated from the uterine exudate, the results of a bacteriological examination following abortion are of little value.

The subsequent breeding history of 23 of these mares is available. Of these, only 9, or 39.5 percent, became pregnant during the first breeding season following abortion.

CLINICAL EXAMINATION OF MARES AFTER FOALING.

The practice of breeding the mare on the first heat period, that is the 9th day, following parturition, is almost universal. The only exception has been when it was evident that the genital tract of the mare was abnormal. Examination of the genital tract of mares after foaling revealed that many were not in a condition favorable to conception, altho they showed no marked external evidence of abnormality. Clinical and bacteriological examinations were made of the genital tract of mares after parturition, for the purpose of determining if it were possible to differentiate between those that were progressing physiologically and those that were not making a normal recovery.

During the first three days after parturition it was usually impossible to determine with certainty from a visual examination of the vagina and cervix whether the prognosis would be favorable or unfavorable. From the fourth to the ninth day rather definite and valuable information was acquired. The great majority of cases could on clinical examination alone be accurately diagnosed during this period. In general, the clinical picture of the genital tract of the mare that has foaled recently, observed between the first and ninth day, varied sufficiently to justify the clinician in making the three following classes:

1. Cases in which the changes are unmistakably physiological.

2. Cases in which the changes are unmistakably pathological.

3. Cases that are intermediate between groups 1 and 2.

In group 1 involution is prompt and complete. The uterine mucosa usually is dry and covered with a dry, gummy residue which has a grayish or reddish-brown tint or, less often, is decidedly black and tenacious. The visible mucous membrane of the cervix and vagina is reddish pink, soft and relaxed, and there is little or no evidence of excretion or lochia around the cervix or in the vagina. Some days later the residue in the uterus tends to liquefy and is passed out so gradually as to be hardly noticeable externally. By the 7th to 10th day the genital tract appears normal, the visible membranes are of a soft, velvety texture, relaxed, have a healthy pink color and are slightly moist.

Based on the results of the bacteriological examinations which, to a certain extent, have a correlation with the clinical picture, it appears that in a small percentage of cases microorganisms do not reach the cervix and uterus following parturition. Such cases are examples of strictly normal birth followed by perfect physiological involution of the uterus and the whole genital tract. The general physical condition of the mare, age, number of foals produced and the conformation of the pelvic region are factors that have a significant bearing for or against bacterial invasion of the genital tract.

As a general rule the mares in group one should receive no treatment and will be ready for service on the first heat period, 9th day.

In the second group involution usually is delayed, the amount of residue in the uterus is pronounced and varies from muco-pus to a liquid, flocculent, purulent discharge. The color varies from light yellow to grayish-brown or brownish-red. Because of the relaxed, atonic condition of the genital tract the uterine excretion is not expelled promptly but tends to accumulate in the uterus and vagina. The exudate is found to be teeming with microorganisms which may easily become established on the vulnerable mucosa of the cervix and uterus, setting up inflammatory processes that are difficult to overcome. Practi-

cally all cases classified under group 2 requires treatment, and the mare is almost never sufficiently recovered by the ninth day to justify admittance to service. From the progress of the case during the next 18 days it can usually be determined whether the mare may be bred on the 27th day or at a subsequent heat period, or should be held over to the following year.

In the third or intermediate group involution is not so prompt as in group one. The uterine residue is more often decidedly mucous in character and considerable in amount, sometimes of a dark dirty brown or brownish-gray color. It tends to accumulate in the uterus and in the vaginal cavity, and is expelled more or less continuously or at intervals, in considerable amount, often soiling the external genitals and adjacent parts. The cervix is as a rule not well contracted, and the membranes are slightly swollen, red and glistening, showing evidence of the beginning of inflammation. Many such cases make a favorable change from the fifth to the ninth day and require no attention. Others grow progressively more abnormal, the excretion becomes muco-purulent in character and the membranes show decided evidence of inflammation.

In the intermediate cases the evidence of congestion and the amount of uterine excretion are more pronounced than may correctly be considered strictly normal. On the other hand, the deviation from the normal is not sufficiently marked to warrant making an unfavorable prognosis nor to justify other than palliative treatment.

The procedure to be followed for mares in group three must be determined for each individual. A bacteriological examination of the uterine excretion taken thru the cervical canal gives information of value, especially if made from the 6th to 9th day; however, the appearance of the genital tract on clinical examination is all important.

The clinician who thru training and experience, has become able to differentiate between the physiological and pathological processes of the genital tract of mares that have foaled recently holds the key to the situation.

A small percentage of mares that passed thru a normal par-

turition with no apparent bad after effects will fail to conceive when bred and later show streptococcic cervicitis and metritis. Some that conceive may abort or give birth to a diseased foal. In such cases there are two possibilities; first that infection was present in the genital tract when the mare was bred or was introduced at the time of service.

The three groups described above include only those cases in which parturition, so far as could be observed, was perfectly normal: An easy uneventful delivery with prompt passage of the placenta intact. There are many cases of parturition that are observably difficult to a minor or major degree. They constitute in the main conditions varying from delayed parturition to actual cases of complicated dystocia. The dystocia is usually fetal but may in some instances be maternal. In all of the wide variety of causes of difficult delivery the usual result is some injury to the genital tract of the dam. Injuries vary from slight bruises and lacerations to those of a more extreme and severe nature. All of these, but particularly severe bruising of the mucosa and subadjacent tissue of the vulvo-vaginal canal and laceration of the lips of the vulva, especially at the upper commissure, decidedly predispose the genital tract to bacterial invasion. Unless some corrective measures are applied the bacteria may become established in the genital tract and cause severe inflammatory changes.

In observing brood mares and in studying their breeding records two extremes are encountered. First there is the mare in poor physical condition that has produced several foals, that suffered some minor injury at time of delivery, that lacked tonicity of the genital structures and that showed an abnormal amount of excretion following parturition. Such a mare when bred on the ninth day following foaling may conceive and in due course give birth to a normal foal. An example of the other extreme is the maiden mare which conceived the first season that she was bred but later aborted. The fetus upon examination was found to be infected with streptococci and the mare developed streptococcic metritis following abortion.

The extremes are of interest, one showing how effective the

natural protective and recuperative powers of nature are under certain conditions, while in the other instance the results demonstrate how completely inadequate the natural protective forces of the body may be in their action against the invasion and multiplication of microorganisms. It is a mistake for anyone to proceed on the assumption that because one individual mare, not in the best of condition and receiving no special attention, stands out in her record of production that a majority will prove equally resistant and consistently productive.

Rather detailed observations have been made on a small group of brood mares during the last four years. The practice followed was to examine every mare that had foaled, one or more times between the first and ninth day following parturition. Unless the mare was normal by the ninth day, so far as bacteriological and clinical methods could determine, she was not admitted to service. For the four years the number of services per pregnancy for the group has been slightly less than two. The percentage of mares in foal has ranged from 60 to 80. This percentage includes all brood mares on the farm. Some were not bred because of temporary unsoundness. During the four foaling seasons covered by the record, forty-five foals were born. There were no serious cases of difficult parturition and all foals were healthy at birth. Twenty-eight fetal membranes from this group of mares were examined in detail and all were found normal. Forty-four foals were weaned. One died at age of 60 days from atrophy and degeneration of the liver, the cause of which was unknown, no infection was found on post-mortem. One mare aborted during her first pregnancy. One mare retained a small piece of afterbirth in the non-pregnant horn. Examination of the membranes showed no evidence of disease. Streptococcic metritis developed and the mare was not bred that season. She was bred the following year and conceived.

It has been definitely established that infections in the new born are often antepartum in origin. Therefore, to undertake effective preventive measures for streptococcic infection it is absolutely fundamental that the condition of the genital tract of the mare be known before breeding. This applies both to the

mare barren during the preceding year and to the mare that has foaled. It is evident however that at no time during the life of the brood mare is she so exposed and susceptible to infection of the genital tract as during the first ten days following parturition.

The prevention of non-specific cases of abortion and the prevention of streptococcic infection of the new born is to be undertaken previous to breeding. If precautionary measures are taken the number of mares that fail to conceive, the development of streptococcic metritis, abortions from streptococcic infection and diseases of the new born resulting from prenatal infection will be reduced.

DISCUSSION

It must be remembered that in the present work *S. abortivo-equinus* infection was not a primary object of study. This is a specific infection for which adequate control measures have been worked out and put into general practice.

In the examination of fetuses streptococci were found responsible for the abortions of 23.4 percent of the cases. It is probable that many more of the abortions were due to streptococcic infection. It is known that infection may be present in the fetal membrane and not present in the fetus. In many instances the membranes were not available and in others their condition rendered a satisfactory examination impossible. *E. coli*, staphylococci, and *S. equirulis* were found in a small number of fetuses but these organisms are of only slight importance in the problem of equine abortion.

Many cases of abortion occur in which no infection is detectable. These were observed both as isolated cases and as quite severe outbreaks. No definite information as to the etiology of these outbreaks has been gained. The results of attempts to transmit the condition by experimental inoculations indicate that no infectious agent is responsible. A consideration of the many possible factors that may have been responsible suggests that in at least two of the outbreaks the feed was involved. In others

there was no indication that the feed was at fault. In these latter cases the number of abortions and the nature of the outbreaks pointed to an infectious agent as the cause. However, since no microorganisms were isolated and transmission experiments were negative, it seems not justifiable to conclude that the abortions were due to an infection. There is, of course, no proof that the different outbreaks had a common cause and certain observations tend to indicate that they were etiologically distinct. In spite of this fact they present many points of similarity and in the writers opinion they were due to some physiological disturbance or to improper nutrition.

It was found that *S. equirulis* was responsible for the death of 39.8 percent of all the foals examined. The evidence accumulated in the study of foals that died of the infection definitely indicates that the disease in young foals is usually of prenatal origin. It was definitely established that certain field causes of the infection, which resulted in abortion or the birth of dead foals, were of prenatal origin. However, experiments to produce the disease as a prenatal infection failed. The infection does not persist in the mare and cause the death of successive foals or set up inflammatory changes in the genital tract that result in sterility. A mare rarely has two foals that die of this infection and after giving birth to a foal infected with *S. equirulis* she recovers as rapidly as a mare that has given birth to a normal foal. Because the method of transmission of the disease is unknown, it is impossible to suggest a method of prevention and control based on definite information.

It has been established beyond reasonable doubt that *S. equirulis* infection in older foals usually is postnatal in origin and is brought about thru the invasion of the blood stream by strongylus larvæ. It is suggested that an analogous condition may exist in the pregnant mare, sometimes terminating in prenatal infection of the fetus with this bacterium. Practically all brood mares are constantly exposed to strongylus infestation and take in large numbers of the parasites. The larvæ penetrate the mucosa of the intestine, and undoubtedly take with them large numbers of microorganisms. *S. equirulis* has been shown

to be present in the alimentary tract of the horse. Granting that these microorganisms are carried into the blood stream of the pregnant mare, it is possible that they find their way to the uterus and invade the fetus. Two of the most destructive genital infections known, Bang's disease of cattle and contagious abortion of mares, usually are contracted by way of the alimentary tract and the organisms reach the uterus thru the general circulation.

In streptococcic infection of foals the prenatal origin of the disease is more obvious than in *S. equirulis* infection. The relation of streptococcic infections in foals to disease of the genital organs of the mare is readily perceived. Streptococci are found in many aborted fetuses. In foals that die in the first few days after birth, not only does the history indicate prenatal infection, but in many instances the fetal membranes present gross evidence of disease. In some cases it is possible to isolate streptococci from the diseased areas of the placenta. Certain mares produce foals year after year which die of streptococcic infection. It has been demonstrated that the genital tract of a mare that has given birth to a normal foal is quickly invaded by streptococci following parturition. In many mares these organisms are able to establish themselves and produce pathologic changes. It is in the mare that has foaled recently that streptococcic infections of the genital tract originate. Dystocia, conditions which predispose to delayed parturition, and major and minor injuries to the genital tract during delivery, are factors in increasing the susceptibility of the mare to streptococcic infection. The primary action of streptococci in breeding diseases is to produce metritis which results in sterility. When a mare infected with these organisms conceives the result usually is abortion or a diseased foal.

The control and prevention of prenatal streptococcic infections, as well as control of streptococcic abortion, depend upon the breeding of only those mares whose genital organs are sound and free of disease; and the observation of strict sanitary precautions at the time of service. Every possible precaution should be taken to prevent the introduction of bacteria into the genital

tract of the mare. This is especially important soon after the mare has foaled.

SUMMARY AND CONCLUSIONS.

1. Streptococci were found in aborted fetuses more frequently than any other microörganism.

2. *S. equirulis*, *E. coli*, and staphylococci were found occasionally in aborted fetuses but were of minor importance.

3. Many instances of apparently non-infectious abortion were observed. These occurred both sporadically and as well-defined outbreaks. The cause of the majority of these cases is unknown.

4. *S. equirulis* was found responsible for the death of a larger number of foals than any other microörganism. Streptococci, also, were responsible for the death of many foals. *E. coli* and *Corynebact. equi* were found occasionally in diseases of foals.

5. In many instances diseases of foals were prenatal in origin.

6. *S. equirulis* was rarely found in the genital tract of the dam of a foal that had died of this infection. This bacterium has no tendency to become localized permanently in the genital tract.

7. *S. equirulis* was present almost constantly in the mouth and pharynx of the normal horse.

8. Streptococci invade the uterus of the mare shortly following parturition. Sometimes they become established and produce genital infection.

9. Thru careful observation of mares that have foaled it is possible to determine whether a normal recovery is being made, and by proper care and treatment to prevent many cases of genital infection.

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IS THERE A FILTERABLE VIRUS OF ABORTION IN MARES?

Since the preparation of the section on apparently non-infectious abortion, Kentucky Bulletin 333, pages 297 to 301, was prepared, some observations have been made that are of great interest in that connection. In December, 1932, and the first three months of 1933, a severe, clear-cut outbreak of abortion that resembled closely the apparently non-infectious abortion previously studied, occurred on a farm near Lexington. Of 32 pregnant mares 24 aborted. The period of gestation of the mares that aborted was between the seventh and tenth months, the great majority about the eighth month. The clinical condition of the mares before and after abortion, the promptness with which the majority of them returned to normal, the absence of cultivable microorganisms, and the pathology of the fetal tissues resembled those conditions observed in previous outbreaks. The results of animal inoculations, however, were entirely different. Four pregnant mares were used in inoculation experiments:

No. 1. A mare in the eighth month of gestation was injected intravenously with 10cc of thoracic fluid and subcutaneously with 5cc of stomach fluid of one of the aborted fetuses. On the same day she was fed the exudate from the fetal membranes. Two weeks later she was fed scrapings from the fetal membranes of another aborted fetus from the same farm. Twenty-eight days after injection she aborted a fetus showing the same pathological changes as were observed in the fetus from which the material for injection was derived. Bacteriological examination of the fetus was negative.

No. 2. A mare in the eighth month of gestation was injected intravenously with 10cc of a suspension of ground liver and spleen and fed 50cc of the same material derived from a fetus from another mare in the same outbreak. After 27 days she aborted a fetus which was bacteriologically negative.

No. 3. A mare, gestation period not known, was injected intravenously with 12cc of a mixture of ground liver and spleen and stomach fluid derived from the fetus from a third mare in

the same outbreak. She was fed 30cc of the same mixture. At the time of writing, 50 days later, she is still pregnant.

No. 4. A mare in the seventh month of gestation was injected intravenously with 10cc of mixed stomach and thoracic fluid, scrapings from membranes, ground liver and spleen of a fetus from the same farm. Before injection the material had been passed thru porcelain filters known to be impervious to bacteria. She was fed 3cc of the same filtrate. After 32 days she aborted a fetus which was negative on bacteriological examination.

Out of four experimental pregnant mares injected and fed material from aborted fetuses, three aborted. Two received unfiltered, and one, filtered material. The period of incubation was practically the same in all cases, therefore the results would strongly indicate the presence of an infectious agent in the inoculum. Since only one mare was inoculated with filtrate, the result cannot be accepted as final that the abortion was directly due to the action of a filterable agent. However, the mare inoculated with filtrate was, so far as could be determined, healthy before exposure and was not either before or following inoculation, in contact with other mares that aborted. Extensive bacteriological studies failed to reveal the presence of cultivable microorganisms. The results may be taken as a tentative indication that in this particular outbreak the abortions were caused by an infectious agent and that the infectious agent is filterable.

